Primary and bacterial production in lakes: are they coupled over depth?

Michael L.Pace and Jonathan J.Cole

Institute of Ecosystem Studies, Box AB, Millbrook, NY 12545, USA

Abstract. The coupling of primary and bacterial production over depth was examined in three lakes which differed greatly in vertical patterns of primary productivity. We measured bacterial production, chlorophyll and light, and estimated primary production in Paul Lake (Gogebic County, Michigan) and Crystal and Trout lakes (Vilas County, Wisconsin) during the summer stratification period (May–September 1991). Bacterial productivity was measured using the [^{3}H]leucine incorporation method and primary productivity estimated from measured photosynthesis–irradiance relationships. Three distinct vertical patterns were observed. In Paul Lake, bacterial production was highest at the interface between the aerobic and anaerobic layers, well below the depth of maximum primary production. In Crystal Lake, bacterial production was uniform with depth, although primary productivity was highest in the hypolimnion. In the largest lake, Trout Lake, primary and bacterial production tended to co-vary with maximum rates of both processes occurring in the metalimnion. Overall, bacterial productivity was poorly related to contemporaneous primary production in the three lakes, suggesting that other factors, such as nutrient recycling, phytoplankton loss rates and allocthonous loading, determine patterns in the depth distribution of bacterial productivity.

Introduction

Bacterial metabolism in aquatic ecosystems has long been viewed as being tightly coupled to primary production (Cole, 1982). This notion is mechanistically reasonable because algal production provides labile substrates and nutrients which support bacterial growth. Comparisons of bacterial and algal production among systems support the concept of this tight coupling. For example, planktonic bacterial production in the surface waters of lakes and marine systems is positively related to primary production (Cole *et al.*, 1988). While bacteria appear to derive much of their carbon from phytoplankton, these two groups also compete for nutrients so that increases in bacteria with primary production may simply reflect the common response of bacteria and phytoplankton to nutrient loading (Currie, 1990). In either case, primary production explains much of the variation in bacterial production among ecosystems. Within ecosystems, the regulation of bacterial productivity seasonally and with depth is less certain. A number of factors, including temperature, dissolved organic carbon, primary production, phytoplankton community structure, grazers, viruses and nutrients, are probably significant, but there are, as yet, few generalizations concerning the relative importance of these factors (Ducklow and Carlson, 1992).

Given the variety of processes that influence bacterial productivity, examining variation in this process across gradients can help isolate potential regulatory factors. In this study, we investigate the relationship between phytoplankton primary production and bacterial secondary production over depth in three lakes. We ask if vertical patterns of bacterial production correspond to vertical patterns of primary production. If bacterial production is closely related to

primary production, one could infer that bacterial productivity is driven by the release of organic carbon and the production of phytoplankton-derived detritus that results from current primary production. If bacterial production is not closely related to primary production, then nutrients, other carbon sources and/ or time lags between primary production and bacterial production are likely to be more important. To address these questions, we examined three temperate lakes with similar volumetric rates of primary production, but differing in vertical patterns of primary production. These lakes were studied during the period of late May to early September when temperature stratification results in stable vertical distributions of primary production.

Method

Study sites

Trout and Crystal Lakes are located in Vilas County, Wisconsin, and are part of the Northern Lakes LTER program (Magnuson *et al.*, 1990). Paul Lake is within the University of Notre Dame Environmental Research Center in Gogebic County, Michigan, and part of an ongoing experimental lakes study (Carpenter and Kitchell, 1993). Primary production has been measured in each lake for a number of years: Paul, 1984–present; Trout, 1985–present; Crystal, 1986–present. All three systems are oligo- to mesotrophic with summer epilimnetic chlorophyll concentrations of the order of 4 $\mu g\, l^{-1}$ in Paul, 1–2 $\mu g\, l^{-1}$ in Crystal and 2–3 $\mu g\, l^{-1}$ in Trout (Adams *et al.*, 1990; Carpenter *et al.*, 1993). Paul Lake is a small (1.5 ha) brown-water system with a summer Secchi depth of 4 m and an anaerobic hypolimnion. Primary productivity is restricted to the upper 5–6 m of the water column and about equally divided between the epilimnion and metalimnion (Carpenter *et al.*, 1993). Crystal is a moderate size (37 ha), clear-water lake (summer Secchi depth 8 m) with an aerobic hypolimnion and primary production occurring throughout the water column (Adams *et al.*, 1990). Trout is a large lake (1608 ha) with a summer Secchi depth of 5 m (Adams *et al.*, 1990). The hypolimnion remains aerobic all summer in the two major basins of the lake.

Sampling

We sampled each lake at bi-weekly intervals from late May to early September in 1991 to obtain a total of eight depth profiles from each system. At each sampling, temperature and oxygen were measured with a YSI model 57 meter. Water was sampled from seven depths whose selection was based on the temperature and oxygen profile, and our prior knowledge of primary production in the three lakes. Water samples were taken by peristaltic pump. Samples for bacterial productivity were pumped from selected depths into 17-ml screw-cap centrifuge tubes. Tubes were flushed several times and filled so that no air bubbles were trapped after closure. Six replicate samples were taken at each depth. Tubes were held in the dark at *in situ* temperature until assay (within 1–2 h of sampling). Samples were also taken at each depth using the pump for

chlorophyll and water chemistry. Chlorophyll samples were filtered through a GF/F filter, frozen and subsequently extracted in methanol. Chlorophyll was determined fluorometrically with corrections for pheopigments (Holm-Hansen and Riemann, 1978).

Measurement of bacterial production

Bacterial production was measured using the [³H]leucine incorporation method (Kirchman, 1993). [³H]Leucine was added to 17-ml centrifuge tubes at a final concentration of 17 nM (sp. act. 60 Ci mmol^{-1}), found in previous experiments to be saturating in these lakes. The samples were incubated at *in situ* temperature for 45 min. Samples were extracted in 5% trichloroacetic acid (TCA) by combining the entire 17 ml sample with 2 ml of 50% TCA. Time-zero controls were made by adding 100 μl of a 1 mM solution of unlabeled leucine and pouring the sample immediately into 2 ml of 50% TCA. One control was treated in this manner for each depth sampled. All samples were heated at 85°C for 30 min, cooled to room temperature and then filtered through 0.45 μm HA Millipore filters at low vacuum (<200 mm Hg). Extraction tubes were rinsed with 2 ml of ice-cold 5% TCA and the rinse poured through the filter. Filters were subsequently rinsed twice with cold 5% TCA, once with cold 80% ethanol and once with distilled water. Filters were placed in a scintillation vial and dissolved with 1 ml of ethylene glycol monomethyl ether. After at least 24 h, a scintillation cocktail (Scintiverse BD) was added and radioactivity was determined with a liquid scintillation counter.

Bacterial productivity was calculated by converting our measure of moles of leucine incorporated day^{-1} to μg C l^{-1} day^{-1} using a conversion factor of 1.546 kg C mol^{-1}, following the assumptions and estimates of Simon and Azam (1989). In previous time course studies, we determined that the uptake of leucine was linear for at least the first 2 h, in agreement with the results of others (Simon and Azam, 1989; Chrost, 1990). We also determined for surface and deep-water samples from each lake that the rate of [³H]leucine incorporation was maximal at the concentration added (17 nM).

The rate of [³H]leucine incorporation is affected by the concentration of added [³H]leucine, by existing intracellular pools, the synthesis of new leucine during the incubation and by the extracellular concentrations of leucine in the environment (Chrost, 1990). To estimate this effect, we conducted a number of isotope dilution experiments in which the [³H]leucine was diluted by additions of unlabeled leucine (Moriarity, 1986; Chrost, 1990). We added [³H]leucine (17 nM) and unlabeled leucine at final concentrations of 0, 8.5, 17, 34 and 68 nM to a series of samples. Experiments were carried out for surface and deep samples from each lake. Using this method, the pool of unlabeled substrates diluting the radiolabeled leucine was estimated by plotting the inverse of the disintegrations per minute (d.p.m.) incorporated against the concentration of cold leucine added. The *X*-intercept of the data fitted by a least squares regression is equal to the concentration of radiolabeled leucine added plus the pool of unlabeled substrates diluting the radiolabeled leucine. We also measured

the concentration of L-leucine directly on one occasion in the surface and deep water of each lake with high-performance liquid chromatography (Mopper and Lindroth, 1982).

Estimation of primary production

Primary production was estimated following the method of Fee (1973a,b). Our general approach was to measure chlorophyll and, using measured photo-synthesis–light relationships for each lake, calculate primary production at the same depths where bacterial production was measured. Specific aspects of the estimation varied among lakes as a result of how the uptake of $^{14}CO_2$ by phytoplankton was measured.

In Paul Lake, ^{14}C uptake was measured *in situ* at a series of depths on four dates in 1991 (S.R. Carpenter *et al.*, unpublished data). We used these data and simultaneous measures of light at depth to establish a photosynthesis–irradiance (P–I) relationship (Platt *et al.*, 1980). In addition, weekly light profiles taken with a LiCor meter (spherical quantum sensor) were used to estimate the extinction coefficient. On the dates when bacterial production was measured, we used surface light data to estimate light at each depth sampled. The specific rate of primary production ($P_b = \mu g\ C\ l^{-1}\ h^{-1}\ chl\ a^{-1}$) was estimated from the P–I curve and converted to an estimate of productivity ($\mu g\ C\ l^{-1}\ h^{-1}$) using our direct measurements of chlorophyll (described above). The ^{14}C uptake measurements were made to a depth of 1% light. We assumed that primary production was negligible below this depth.

In Crystal Lake, P–I experiments were conducted approximately every other week by the Northern Lakes LTER program. Integrated water samples taken from the epi-, meta-, and hypolimnion, and ^{14}C uptake measured at a series of irradiances [as described in Adams *et al.* (1990)]. We used these data to establish a P–I curve for each stratum of the lake over our sampling period. The light extinction coefficient varies with depth in Crystal because the accumulation of chlorophyll in the hypolimnion increases light extinction in deeper waters. We fitted two separate regressions to estimate extinction: one for the combined epilimnion and metalimnion data, and one for the hypolimnion. Surface light data collected daily by the Northern Lakes LTER program were averaged for the period from 9 a.m. to 3 p.m. to estimate average light conditions for the dates of our sampling. Primary production at each specific depth was then calculated using the extinction coefficients, the stratum-specific P–I curves and chlorophyll.

The procedure for estimating primary production in Trout Lake was similar to that for Crystal. P–I experiments were only available for the epilimnion of Trout in 1991, so we used P–I data for the metalimnion and hypolimnion from 1989 and 1990 to establish P–I curves from these strata. The other difference between Crystal and Trout was that light extinction was uniform with depth in Trout, so a single regression was suitable for describing the relationship between light and depth.

Results and discussion

Isotope dilution and estimation of bacterial production

Calculation of the rate of bacterial production is dependent on the specific activity of the radiotracer prior to product formation (Karl, 1982). In the case of leucine, which is a nearly constant percentage of total protein (Simon and Azam, 1989), once intracellular and extracellular pools of [³H]leucine equilibrate the rate of protein formation should be directly related to the rate of [³H]leucine incorporation into protein if no other leucine is present. Extracellular and intracellular leucine (hereafter the diluting pool) reduce this rate. We estimated this reduction or 'dilution' of the radiolabel in two ways. First, using standard isotope dilution experiments, we found that in the surface waters of the three lakes the concentration of the diluting pool was very low (Table I). In two cases, our estimate of [³H]leucine plus the diluting pool (= X-int in Table I) was less than the amount of [³H]leucine added and in one case was slightly greater. Thus, little or no unlabeled leucine was present or synthesized during the incubations. In the deeper waters of the three lakes, dilution was significant, ranging from 1.3 to 2.8 times the added leucine (Table I). The second approach was to measure the environmental concentration of leucine directly. These direct measurements do not assess the amount of intracellular leucine diluting the added [³H]leucine, potentially leading to an underestimate. Some fraction of the small peptides free in the water, however, may be measured as their constituent amino acids in the analysis, leading to a potential overestimate of the actual concentration of leucine (C.Lee, personal communication). In any case, using the direct measures of leucine, we find that dilution is ~2-fold in the surface waters and 3- to 4-fold in deep waters (Table I). The two approaches yield the same pattern of lower dilution in the surface and higher dilution in the deeper layers of these lakes, but differ in the estimated magnitude of dilution.

We used a factor of two for isotope dilution to calculate bacterial production for all lakes and depths. This factor agrees closely with the estimates of other

Table I. Dilution of ³H-labeled leucine as estimated by isotope dilution experiments and by direct measurement of the leucine pool size. Isotope dilution experiments were conducted in July 1991 in Trout and Crystal and in September 1991 in Paul. All samples for direct measurement of leucine were taken in September 1991. Absolute value of the X-intercept (= X-int) represents the concentration of added radioactive leucine plus diluting pool. Dilution factor 1 calculated from the absolute value of the X-intercept (= X-int). n is the number of observations and r^2 the total variance explained by least squares regression for each dilution experiment. Dilution factor 2 calculated from the concentration of L-leucine measured directly by HPLC (see the text)

Lake	Depth (m)	[³H]Leu (nmol l⁻¹)	X-int (nmol l⁻¹)	Dilution factor 1	n	r^2	Measured L-leucine (nM)	Dilution factor 2
Trout	0	17.2	18.86	1.11	14	0.75	17.2	2.00
	28	17.2	32.62	1.92	14	0.70	49.6	3.88
Crystal	9	17.2	6.60		14	0.91	21.9	2.27
	19	17.2	21.69	1.28	13	0.84	35.9	3.09
Paul	0	17.2	6.72		15	0.98	15.4	1.90
	9	17.2	47.36	2.79	15	0.50	40.6	3.36

investigators (Simon and Azam, 1989; Chrost, 1990). A 2-fold dilution factor either overestimates or is nearly correct for the surface waters of our study lakes and either underestimates or is nearly correct for the deep waters (Table I). The absolute magnitude of our estimates of [³H]leucine incorporation are, therefore, uncertain by a factor of ~0.5–2. Most of the arguments concerning relationships with primary production that we develop below are qualitative and not affected by a 2-fold uncertainty in absolute rate estimates.

Volumetric and areal rates of primary and bacterial production

Average volumetric and areal rates of primary production were highest in Trout Lake and lowest in Paul Lake (Table II). The highest average volumetric rates of bacterial production were observed in Paul and the lowest rates in Trout, a pattern opposite to the trend in volumetric primary productivity (Table II). Areal bacterial production estimates were remarkably similar, differing by only a few percent among the three lakes (Table II).

   Bacterial productivity, as a percentage of primary productivity, was highest in Paul and lowest in Trout. The percentages in each lake were lower than the volumetric average of 20% and the areal average of 30% reported by Cole *et al.* (1988) for a large number of freshwater and marine systems. There was considerable scatter in the relationship of Cole *et al.* (1988), and the average values for Paul, Crystal and Trout do not fall outside the 95% confidence limits for individual predictions. Nevertheless, the consistently lower estimates we derived suggest either that our method using [³H]leucine incorporation yields a lower estimate of bacterial productivity or that bacterial productivity was lower in our study lakes than in most systems. The data summarized by Cole *et al.* (1988) do not include any measurements made using the leucine method, although direct comparisons between the leucine and thymidine method suggest that these techniques give similar estimates (Kirchman, 1992). There is, however, no ecological basis for assuming that bacterial productivity in our study lakes should be particularly low, and so we tentatively conclude that our method of measuring [³H]leucine incorporation yields a lower estimate of bacterial production relative to other studies.

Patterns with depth

Vertical patterns of primary and bacterial production within each lake were

Table II. Comparison of average volumetric and areal rates of primary (PP) and bacterial (BP) production in the three study lakes. Note that volumetric rates are for depths above the 1% light level. Hourly rates were converted to daily rates using factors of 12 and 24 for PP and BP, respectively

Lake	Volumetric (μg C l^{-1} day^{-1})			Areal (mg C m^{-2} day^{-1})		
	PP	BP	BP:PP (%)	PP	BP	BP:PP (%)
Paul	31.7	3.81	12	197	50	25
Crystal	35.4	2.72	8.7	637	51	8.0
Trout	46.8	2.38	5.1	839	48	5.7

fairly similar throughout the May–September sampling period. In Paul, primary production was either uniform in the upper 3 m or showed a metalimnetic maximum (Figure 1). There was always a peak in bacterial productivity near the top of the anaerobic zone at a depth of 6–7 m. This appears to be a site of intense microbial activity in Paul Lake, but due to low light levels, there is little or no photosynthesis by phytoplankton at this depth (Figure 1). Crystal Lake was characterized by a deep-water maximum in primary production, but bacterial production in this lake was essentially uniform with depth (Figure 1). Primary production in Trout was highest over the depth interval 7–10 m (Figure 1). These depths were at the base of the mixed layer and the upper portion of the thermocline. The depth distribution of bacterial production was often similar to that of primary production (Figure 1).

An interesting feature of Figure 1 and all the vertical profiles was the relative magnitude of the variation in bacterial and primary production over depth. There was often a $\geq$10-fold range in rates of primary production. This variability was not just the result of light extinction. For example, in the Crystal Lake profile illustrated in Figure 1, primary production varied from 9 μg C l^{-1} day^{-1}
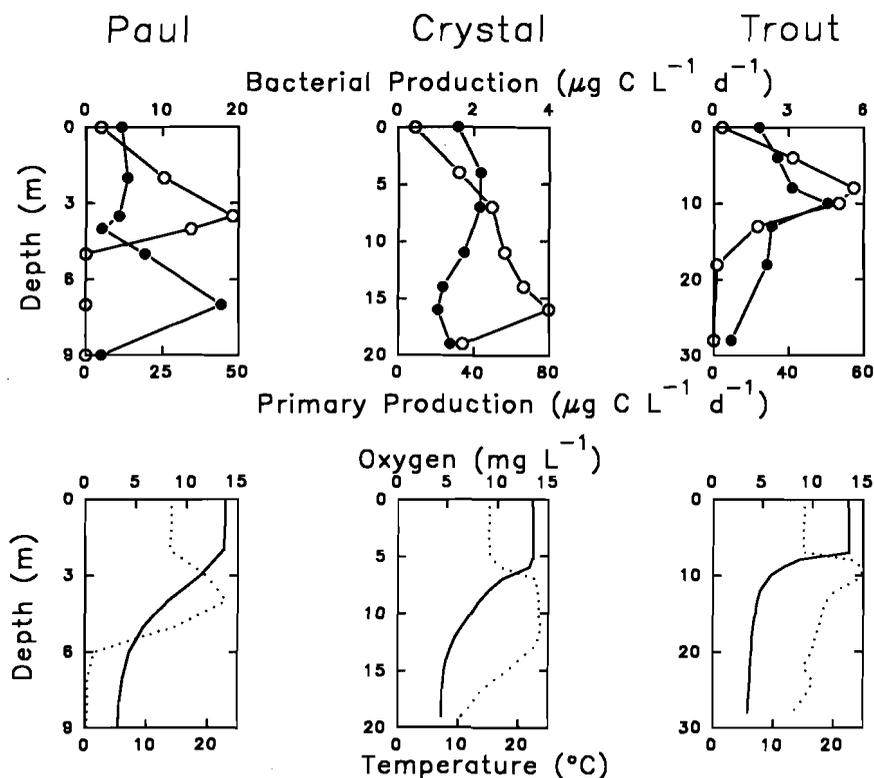

Fig. 1. Typical patterns of primary (○) and bacterial (●) production with depth in Paul (left), Crystal (middle) and Trout (right) lakes from July 1991. Below each productivity panel are profiles of temperature (solid line) and oxygen (dotted line) for the same date.

at the surface to 80 μg C l^{-1} day^{-1} at 16 m. In contrast, the variation in bacterial productivity with depth was modest, as in the illustrated profile for Crystal Lake (Figure 1) where bacterial productivity varied from only 1 to 2 μg C l^{-1} day^{-1}.

While there was a correlation between log-transformed bacterial and primary production ($r = 0.47$, $P < 0.0001$, $n = 148$), the considerable scatter in the data (Figure 2) indicates that bacterial production was not well related to current primary productivity. A qualitative approach to testing the hypothesis that current primary production drives bacterial production is to examine rank correlations for individual profiles in each lake. Here, we ask if the ranks of primary and bacterial production over depth tend to be positively correlated. In Paul and Crystal, there was clearly no tendency for the rank correlations to be positive (Table III). The ranks for all profiles in Trout were positively correlated (Table III) and in three cases these correlations were significant ($P < 0.05$). In Trout, bacterial and primary production did co-vary so that vertical profiles have a similar pattern (Figure 1c), but the actual magnitude of the variability in bacterial production was small. Most of the observations (85%) were in the range of 0–3 μg C l^{-1} day^{-1}, while primary production varied >100-fold (Figure 2).

Implications

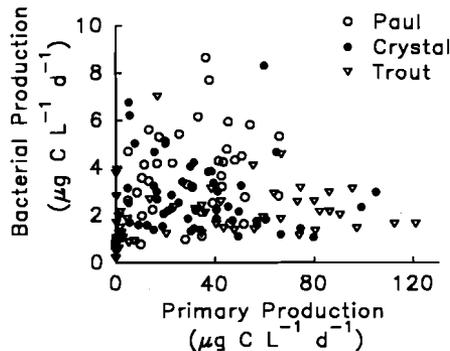The strong associations between bacterial and primary production found for a

Fig. 2. Primary and bacterial production for all dates and depths sampled.

Table III. Rank correlations for 24 individual profiles of primary and bacterial production in the three lakes. This statistic tests qualitatively whether bacterial and primary production tend to be correlated over depth for individual profiles. Numbers of profiles for each lake are indicated for which these variables were negatively correlated, positively correlated, positively correlated and significant ($P < 0.05$)

Lake	Negative r	Positive r	Positive r ($P < 0.05$)
Paul	5	3	1
Crystal	5	3	0
Trout	0	8	3
Total	10	14	4

number of systems (Cole *et al.*, 1988), and seasonally within some systems (e.g. Bell and Kuparinen, 1984; Chrzanowski and Hubbard, 1988), do not hold over depth gradients in the lakes studied here. This uncoupling of bacterial and primary production has important implications for carbon cycling. Rapid coupling between contemporaneous phytoplankton production and bacterial metabolism cannot account for the vertical patterns observed. Bacterial productivity may depend on both current and prior primary production through the simultaneous use of new photosynthate and the degradation of particulate remains of phytoplankton. This result is consistent with recent analyses suggesting that the release of photosynthetically derived dissolved organic carbon is, in general, insufficient to support bacterial production (Jumars *et al.*, 1989; Baines and Pace, 1991). Studies of bloom dynamics in lake, coastal and oceanic systems reveal that increases in bacterial productivity often lag increases in phytoplankton productivity and biomass (e.g. Billen and Fontigny, 1987; Simon and Tilzer, 1987; Ducklow *et al.*, 1992). An additional mechanism uncoupling primary and bacterial productivity is the sedimentation of algal detritus. In this regard, Cole *et al.* (1984) found that in Mirror Lake the decomposition of particulate algal material supported a larger fraction of bacterial production than the release of algal exudates from contemporaneous primary production.

The best association between primary and bacterial production was observed in Trout Lake, which is by far the largest of the three lakes. In a large lake, especially one with a long water residence time and low allocthonous loading, we might expect to observe a closer association between primary and bacterial productivity with depth. There is some support for this conjecture from prior studies. Depth profiles of bacterial production appear to conform with vertical distributions of chlorophyll and/or primary production in large systems such as Lake Biwa (Nagata, 1987) and Lake Michigan (Fahnenstiel and Scavia, 1987; Scavia and Laird, 1987). In many smaller lakes, especially those with anaerobic hypolimnia like Paul Lake, bacterial production is often highest in the metalimnion (e.g. Lovell and Knopka, 1985; McDonough *et al.*, 1986; Bloem and Bär-Gilissen, 1989). In these systems, there appears to be greater temporal and spatial uncoupling of the processes of primary and bacterial production. The same logic may apply in marine systems. In nearshore regions, bacterial and primary production are often unrelated on both horizontal and vertical scales because physical dynamics and interactions with sediments uncouple phytoplankton and bacterial processes (e.g. Pomeroy *et al.*, 1983; Joint and Pomroy, 1987). In oceanic environments with a high degree of vertical stability in temperature and density, primary and bacterial production appear to be more closely related (Ducklow and Carlson, 1992).

Our data suggest that either the phytoplankton-derived carbon utilized by bacteria is more variable than phytoplankton biomass and primary production, or that factors other than primary production determine the vertical distribution of bacterial production during the stratification period. One such factor may be the availability of phosphorus and nitrogen (Currie, 1990). We found no positive relationships between nutrients (NH_4, NO_3, SRP, TP) and bacterial production,

except for a weak relationship with TP (log-transformed data where $O_2 > 1$ mg l^{-1}; $r = 0.15$, $P = 0.07$, $n = 149$). Nutrients may well limit bacteria in these lakes, but if nutrients are reduced to low concentrations, the rate of nutrient recycling, which is difficult to measure, may be the most important correlate to bacterial growth (T.Chrzanowski, personal communication). Prior experiments in Paul Lake reveal that additions of N and P increase bacterial growth and biomass, implying that nutrients, not phytoplankton-derived carbon, most limit bacteria in these lakes (Pace, 1993).

Variability in bacterial production in the three lakes was modest. For example, in Paul, Crystal and Trout lakes, the coefficients of variability (CVs) for bacterial productivity in the photic zone were 0.49, 0.54 and 0.49, respectively. By comparison, CVs for primary production were 0.51, 0.67 and 0.71. The lower variability in bacterial productivity is consistent with comparative studies. For example, Cole *et al.* (1988) observed a much greater range in primary than in bacterial productivities. It is also well known that variation in bacterial biomass is small in comparison with variation in algal biomass among systems (Simon *et al.*, 1992). Together, these observations suggest that there are severe constraints on bacterial communities that limit both increases and decreases in productivity and biomass relative to other components of the system.

The highest rates of bacterial productivity for all three study lakes occurred at the interface between aerobic and anaerobic waters in Paul Lake. Intensive microbial activity is often observed in lakes at this interface. We have also observed that bacterial cell size is larger in the anaerobic waters of lakes (Cole *et al.*, 1993). These observations lead to the hypothesis that the relative significance of bacterial productivity and biomass will be greater in lakes with anaerobic hypolimnia.

We conclude that there are important, but poorly understood, lags between the formation of phytoplankton biomass by primary production and the degradation of phytoplankton productivity by bacterial metabolism. Discontinuities associated with chemoclines are important in determining vertical patterns of microbial activity and may partially explain differences among lakes in rates of bacterial productivity. Processes associated with nutrient recycling are also likely to be significant in determining the vertical profile of bacterial production.

A complete theory of bacterial productivity should explain patterns at several scales, including over depth and across seasons, within as well as among ecosystems. While we have a limited ability to predict bacterial abundance and productivity across gradients of enrichment, few models describe bacterial dynamics or explain similarities and differences in dynamics for a variety of systems. By comparison, there is a rich spectrum of models for phytoplankton production. These are often derived from general physiological responses (e.g. P–I curves) and easily measured variables (e.g. light extinction). Analogous models for bacteria require that the most significant processes driving productivity be identified (e.g. substrate supply, temperature, nutrient availability) and incorporated into relatively simple models that can be widely tested.

Acknowledgements

References

Adams,M.S., Meinke,T.W. and Kratz,T.K. (1990) Primary productivity in three northern Wisconsin lakes, 1985–1987. *Verh. Int. Ver. Limnol.*, **24**, 432–437.

Baines,S.B. and Pace,M.L. (1991) The production of dissolved organic matter by phytoplankton and its importance to bacteria: Patterns across marine and freshwater systems. *Limnol. Oceanogr.*, **36**, 1078–1090.

Bell,R.T. and Kuparinen,J. (1984) Assessing phytoplankton and bacterioplankton production during early spring in Lake Erken, Sweden. *Appl. Environ. Microbiol*, **48**, 1221–1230.

Billen,G. and Fontigny,A. (1987) Dynamics of *Phaeocystis*-dominated spring bloom in Belgium coastal waters. II. Bacterioplankton dynamics. *Mar. Ecol. Prog. Ser.*, **37**, 249–257.

Bloem,J. and Bär-Gilissen,M.-J.B. (1989) Bacterial activity and protozoan grazing potential in a stratified lake. *Limnol. Oceanogr.*, **34**, 297–309.

Carpenter,S.R. and Kitchell,J.F. (eds) (1993) *The Trophic Cascade in Lakes.* Cambridge University Press, New York.

Carpenter,S.R., Morrice,J.A., Soranno,P.A., Elser,J.J., MacKay,N.A. and St Amand,A.L. (1993) Primary production and its interactions with nutrients and light transmission. In Carpenter.S.R. and Kitchell,J.F. (eds), *The Trophic Cascade in Lakes.* Cambridge University Press, New York, pp. 225–251.

Chrost,R.J. (1990) Application of isotope dilution principle to the determination of substrate incorporation by aquatic bacteria. *Arch. Hydrobiol. Beih. Ergebn. Limnol.*, **34**, 111–117.

Chrzanowski,T.H. and Hubbard,J.G. (1988) Primary and bacterial secondary production in a southwestern reservoir. *Appl. Environ. Microbiol.*, **54**, 661–669.

Cole,J.J. (1982) Interactions between bacteria and algae in aquatic ecosystems. *Annu. Rev. Ecol. Syst.*, **13**, 291–314.

Cole,J.J., Likens,G.E. and Hobbie,J.E. (1984) Decomposition of planktonic algae in an oligotrophic lake. *Oikos*, **42**, 257–266.

Cole,J.J., Findlay,S. and Pace,M.L. (1988) Bacterial production in fresh and saltwater: a cross system overview. *Mar. Ecol. Prog. Ser.*, **43**, 1–10.

Cole,J.J., Pace,M.L., Caraco,N.F. and Steinhart,G.S. (1993) Bacterial biomass and cell size distributions in lakes: More and larger cells in anoxic waters. *Limnol. Oceanogr.*, **38**, 1627–1632.

Currie,D. (1990) Large-scale variability and interaction among phytoplankton, bacterioplankton and phosphorus. *Limnol. Oceanogr.*, **35**, 1437–1455.

Ducklow,H.W. and Carlson,C.A. (1992) Oceanic bacterial production. *Adv. Microb. Ecol.*, **12**, 113–181.

Ducklow,H.W., Kirchman,D.L., Quinby,H.L., Carlson,C.A. and Dam,H.G. (1992) Stocks and dynamics of bacterioplankton carbon during the spring bloom in the eastern North Atlantic Ocean. *Deep-Sea Res.*, **40**, 245–263.

Fahnenstiel,G.L. and Scavia,D. (1987) Dynamics of Lake Michigan phytoplankton: primary production and growth. *Can. J. Fish. Aquat. Sci.*, **44**, 499–508.

Fee,E.J. (1973a) A numerical model for determining integral primary production and its application to Lake Michigan. *J. Fish. Res. Board Can.*, **30**, 1447–1468.

Fee,E.J. (1973b) Modeling primary production in water bodies: a numerical approach that allows vertical inhomogeneities. *J. Fish. Res. Board Can.*, **30**, 1469–1473.

Holm-Hansen,O. and Riemann,B. (1978) Chlorophyll *a* determination: improvements in methodology. *Oikos*, **30**, 438–447.

Joint,I.R. and Pomroy,A.J. (1987) Activity of heterotrophic bacteria in the euphotic zone of the Celtic Sea. *Mar. Ecol. Prog. Ser.*, **41**, 1155–1165.

Jumars,P.A., Penry,D.L., Baross,J.A., Perry,M.J. and Frost,B.W. (1989) Closing the microbial loop: dissolved carbon pathways to heterotrophic bacteria from incomplete ingestion, digestion and absorption in animals. *Deep-Sea Res.*, **36**, 483–495.

Karl,D.M. (1982) Selected nucleic acid precursors in studies of aquatic microbial ecology. *Appl. Environ. Microbiol.*, **44**, 891–902.

Kirchman,D.L. (1992) Incorporation of thymidine and leucine in the subarctic Pacific: application to estimating bacterial production. *Mar. Ecol. Prog. Ser.*, **82**, 301–309.

Kirchman,D.L. (1993) Leucine incorporation as a measure of biomass production by heterotrophic bacteria. In Kemp,P.F., Sherr,B.F., Sherr,E.B. and Cole,J.J. (eds), *Handbook of Methods in Aquatic Microbial Ecology*. Lewis Publishers, Boca Raton, FL, pp. 509–512.

Lovell,C.R. and Konopka,A. (1985) Primary and bacterial production in two dimictic Indiana lakes. *Appl. Environ. Microbiol.*, **49**, 485–491.

Magnuson,J.J., Benson,B.J. and Kratz,T.K. (1990) Temporal coherence in the limnology of a suite of lakes in Wisconsin, USA. *Freshwater Biol.*, **23**, 145–159.

McDonough,R.J., Sanders,R.W., Porter,K.G. and Kirchman,D.L. (1986) Depth distribution of bacterial production in a stratified lake with an anoxic hypolimnion. *Appl. Environ. Microbiol.*, **52**, 992–1000.

Mopper,K. and Lindroth,P. (1982) Diel and depth variation in dissolved free amino acid and ammonium in the Baltic Sea determined by shipboard HPLC analysis. *Limnol. Oceanogr.*, **27**, 336–347.

Moriarity,D.J.W. (1986) Measurement of bacterial growth rates in aquatic systems from rates of nucleic acid synthesis. *Adv. Microb. Ecol.*, **9**, 245–292.

Nagata,T. (1987) Production rate of planktonic bacteria in the north basin of Lake Biwa, Japan. *Appl. Environ. Microbiol.*, **53**, 2872–2882.

Pace,M.L. (1993) Heterotrophic microbial processes. In Carpenter,S.R. and Kitchell,J.F. (eds), *The Trophic Cascade in Lakes*. Cambridge University Press, New York, pp. 252–277.

Platt,T., Gallegos,C.L. and Harrison,W.G. (1980) Photoinhibition of photosynthesis in natural assemblages of marine phytoplankton. *J. Mar. Res.*, **38**, 687–701.

Pomeroy,L.R., Atkinson,L.P., Blanton,J.O., Campbell,W.B., Jacobsen,T., Kerrick,K.H. and Wood,A.M. (1983) Microbial distribution and abundance in response to physical and biological processes on the continental shelf of the southeastern USA. *Cont. Shelf Res.*, **2**, 1–20.

Scavia,D. and Laird,G.A. (1987) Bacterioplankton in Lake Michigan: dynamics, controls, and significance to carbon flux. *Limnol. Oceanogr.*, **32**, 1017–1033.

Simon,M. and Azam,F. (1989) Protein content and protein synthesis rates of planktonic bacteria. *Mar. Ecol. Prog. Ser.*, **51**, 201–213.

Simon,M. and Tilzer,M.M. (1987) Bacterial responses to seasonal changes in primary production and phytoplankton biomass in Lake Constance. *J. Plankton Res.*, **9**, 535–552.

Simon,M., Cho,B.C. and Azam,F. (1992) Significance of bacterial biomass in lakes and the ocean: comparison to phytoplankton biomass and biogeochemical implications. *Mar. Ecol. Prog. Ser.*, **86**, 103–110.